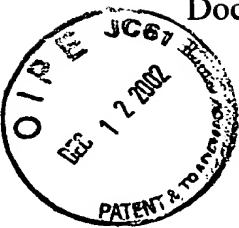


Docket No. 2527-1A



IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:

MARIO TENUTA ET AL.

Serial No. 09/624,098

Art Unit: 1651

Filed: July 24, 2000

Examiner: D. Naff

Title: METHOD FOR THE TREATMENT OF A SOIL  
CONTAINING SOILBORNE PATHOGENS

November 18, 2002

Honorable Commissioner of  
Patents and Trademarks,  
Washington, D.C. 20231

DECLARATION

Sir:

I, George Lazarovits, do hereby declare as follows:

THAT, I am one of the inventors named in the above identified application;

THAT, I am aware of the rejection set forth in the Office Action of July 2, 2002  
wherein the Examiner states that the primary reference of Anderson et al. discloses that  
potato scab disease is known to be controlled by increasing soil acidity;

THAT, I had the following test done to show the effect of reducing soil pH with  
sulfuric acid and the results on potato scab disease, the summary of the test being set forth  
hereinbelow;

**Purpose:**

Nitrite becomes converted to nitrous acid under acid conditions. At about pH 4.3 nitrite and nitrous acid are in equilibrium (50:50). Nitrite has no toxicity to microorganisms such as *Streptomyces scabies*, the bacteria that causes potato scab. Nitrous acid at 0.03 mM however, is lethal. In order to obtain sufficient amounts of nitrous acid it is necessary to lower soil pH for a short time to about pH 4 - 5. This study was carried to determine the effect lowering the pH, in the absence of nitrite, on disease incidence caused by *S. scabies*.

**Materials and Methods:**

Soil known to have high disease pressure for potato scab was collected from a commercial potato field in Ontario in the spring of 2001 and brought to the London research station. An experiment was set up using microplots. Soil (13 kg) was placed in drainage tiles (25 cm deep, 25 cm diam) that had been buried in soil to ground level. The soil pH in some soils was reduced by adding sulfuric acid (45%, obtained from Ethyl Canada Ltd., Sarnia, ON). Acid (7.2 and 16.9 mL) was added to water (390 mL) and the water was then added to soil. The pH was found to be reduced from 6.5 to about 5 and 4, respectively (Figure 1). One week later, a potato seed piece was planted in each tile (four tiles per treatment). The tubers were harvested in the fall and rated for scab on a scale of 0-6 based on the percentage of tuber surface covered with scab lesions where 0 = 0%, 1 = trace to 5%, 2 = 6-15%, 3 = 16-25%, 4 = 26-35%, 5 = 36-60%, and 6 = 61-100%.

**Results and Discussion:**

Soil pH was reduced to 5 and 4 at day zero by the addition of sulfuric acid (Figure 1). Within 2 days however, the pH in acidified soils was up by half a log unit

and at the end of a week a full log unit (Figure 1). Since nitrous acid kills *S. scabies* bacteria within hours the length of time the pH stayed low would have been more than sufficient to control disease. By week 6 the pH was nearly identical in both treatments to the control soil (Figure 1). At harvest we found no effect of pH alterations on the severity of potato scab as compared to the control treatment (Figure 2). Scab severity was near maximum in all treatments. This demonstrates that low pH and acid alone has no effect on disease incidence.

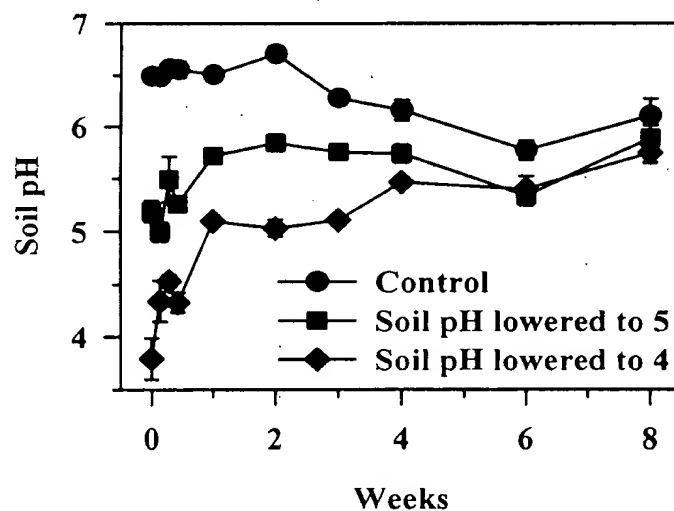


FIGURE 1. Reduction of the pH of a soil from a commercial potato field in Ontario using sulfuric acid. Soil pH was then monitored for 8 weeks. Error bars represent S.E., n = 4.

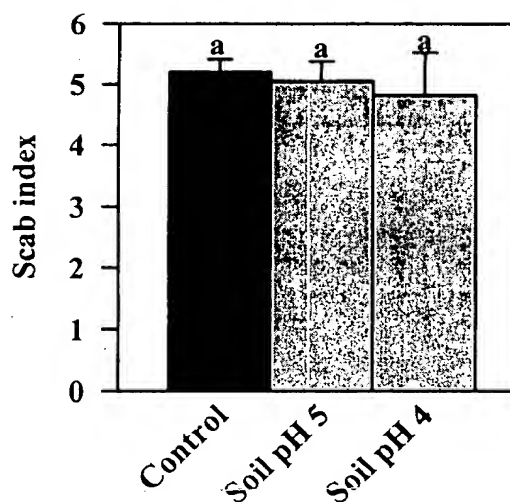


FIGURE 2. Effect of reducing soil pH on potato scab severity. The pH of a soil from a commercial potato field was reduced with sulfuric acid (see Figure 1). Potatoes were planted 1 week later and tubers harvested in the fall were rated for scab on a scale of 0-6 based on the percentage of tuber surface covered with scab lesions where 0 = 0%, 1 = trace to 5%, 2 = 6-15%, 3 = 16-25%, 4 = 26-35%, 5 = 36-60%, and 6 = 61-100%. Error bars represent S.E.,  $n = 4$ . Treatments with the same letter are not significantly different at the 0.05 probability level.

THAT, as shown by this test, the reduction of soil pH by sulfuric acid has no effect on potato scab disease.

FURTHER DEPONETH SAYETH NOT.

  
George Lazarevits

SIGNED this 29<sup>th</sup> day of November, 2002.